

Measurement and comparison of glycyrrhizic acid contents in root of licorice (*Glycyrrhiza uralensis* Fisch.) from different cultivating areas

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Abstract: The samples of licorice (*Glycyrrhiza uralensis* Fisch.) from 14 different cultivated areas were determined by the method of high Performance Capillary Electrophoresis (HPCE) for the contents of glycyrrhizic acid (GA) in root. The results showed that the licorice plants come from various cultivated areas of China has different contents of GA. The GA content of licorice from Zhaodong in Heilongjiang Province is the highest, followed by those from E'tuoke, Chifeng, and Hangjin Banner in Inner Mongolia. Some suggestions for establishing the production base of licorice were put forward based on the study.

Keywords: Glycyrrhizic acid; High performance capillary electrophoresis (HPCE); *Glycyrrhiza uralensis* Fisch.

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Introduction

Licorice (*Glycyrrhiza uralensis* Fisch.) is a perennial herb and widely distributed in the northern, northeastern, and northwestern regions of China. It has strong abilities of drought resistance, cold tolerance, salt and alkali tolerance, checking wind, and controlling sand. As an economic plant, the roots and rhizomes of Licorice are usually used as Chinese herbal medicine (Fu 1989; Liu *et al.* 1989) to treat the diseases such as bronchitis, sore throat, gastroduodenal ulcers, skin inflammation, etc (Sun *et al.* 2000; Lv *et al.* 1999). In addition, licorice can also be employed as a flavour-enhancing agent or a food condiment. Its sale volume is over the other herbs. However, due to predatorily digging roots and rhizomes for a long time, the wild resources of this plant is now facing with exhausting and the ecological environment of the licorice growing areas becoming worsened unceasingly (Wang *et al.* 1999; Rui 1997; Yu *et al.* 1999). Thus, the practice in cultivation of licorice in those areas was performed. The root cultivation technique of Licorice in China was developed by the researchers at the Heilongjiang Institute of Traditional Chinese Medicine in 1962, and since 1980s this technique has been actualized in large scale in Three North-Regions.

Glycyrrhizic acid (GA) is a major active component in licorice root and rhizome for assessment of licorice quality. Licorice from various producing areas is different in quality. This study is aimed at providing scientific basis for evaluating the quality of cultivated licorice and choosing the suitable growing areas. In the study, a high performance

capillary electrophoretic method (Zu *et al.* 2001; Liang *et al.* 1998; Peng *et al.* 1999; Yin *et al.* 1999) was adopted to measure the contents of GA in the cultivated licorice from different producing areas.

Study methods

Preparation of the standard solution

GA standard sample was provided by China National Institute of Drugs Control, A standard sample that weighs 11.2 mg was dissolved with 50% ethyl alcohol in a 50-mL volumetric flask, after then, 2, 4, 6, 8, and 10 mL of solution were drew out of the standard solution and separately placed into a 10-mL volumetric flask, followed by addition of 50% ethyl alcohol.

Buffer solution

Phosphoric acid with a pH value of 5.8 was used as a buffer solution and filtrated with 0.45 μ m micro-filter membrane.

Plotting standard curve

Five standard samples in different concentrations were put into a 270A-HT high performance capillary electrophoresis (from American PE Co.) separately, and each sample was determined three times for using the mean value of the peak area, the peak area was measured as 129 528, 243 501, 479 944, 628 420, and 869 912 mm² individually. When the values of peak area were use as the ordinate axis and the contents of GA as the abscissa axis, the regression equation was obtained as $Y = 4.16 \times 10^6 X - 8.94 \times 10^4$, the correlation coefficient $r = 0.9939$. The standard curve is drawn as Fig. 1. The experimental results showed that there exists a significant correlation between the concentration of standard solutions

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and the peak-area values when concentration of standard solution is in the range of 0.0448-0.224 mg·mL⁻¹, under which condition the GA could be better isolated (Fig. 2).

Determination of GA contents

Preparation of sample solution: The licorice samples were dried at 60 °C , powdered and sieved through 60-mesh-sieve. The dried sample that accurately weighs 1.0000 g, was extracted with adding 80% ethyl alcohol by Soxhlet method for 4 h, after then it was filtered and placed into a 100-mL volumetric flask, followed by addition of 80% ethyl alcohol.

The measurement: Each licorice sample solution was placed into the high performance capillary electrophoresis and determined repeatedly five times. The average value of peak area was used to calculate the GA contents by the standard linear regression equation (Table 1).

Rate of recovery: Fourteen powdered samples of licorice were weighed up accurately and 1.5 mg of standard GA was added to each sample. The licorice sample was determined three times by the above-mentioned method, and the mean value was used to calculate the recovery rate and a 97.5% recovery rate was obtained.

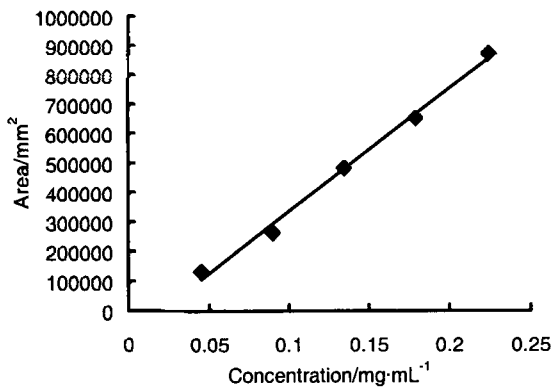


Fig. 1 Calibration of glycyrrhizic acid

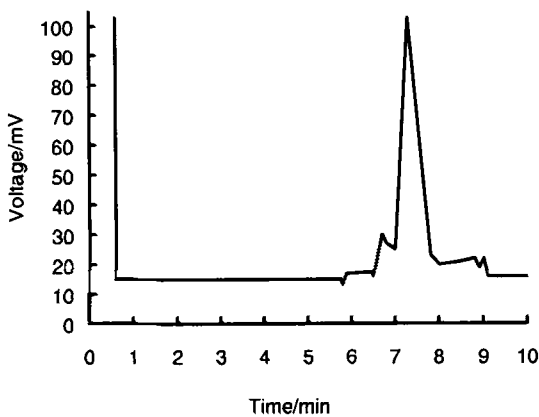


Fig. 2 Electrophoretogram of standard of glycyrrhizic acid by capillary electrophoresis

Table 1. The contents of GA in cultivated *G. uralensis* root from different producing areas

No.	Cultivating area	Content of GA (%)
1	Zhaodong, Heilongjiang	11.41
2	Huanan, Heilongjiang	5.75
3	Zhenlai, Jilin	6.33
4	E'tuoke Front Banner, Inner Mongolia	10.83
5	HangjinBanner, Inner Mongolia	9.52
6	Chifeng, Inner Mongolia	9.36
7	Yanchi, Ningxia	6.35
8	Longde, Ningxia	7.44
9	Minqin, Gansu	7.03
10	Dunhuang, Gansu	6.95
11	Jinta, Gansu	7.22
12	Shaya, Xinjiang	6.04
13	Luntai, Xinjiang	6.56
14	Bachu, Xinjiang	6.38

Results and discussion

The method of high performance capillary electrophoresis to determine the GA content of cultivated licorice has some outstanding characteristics, such as quick determination, good differentiation rate, and high accuracy.

The experimental results showed that the GA content in root of the cultivated licorice was very different between the different growing areas (Table 1). The GA content of licorice from Zhaodong in Heilongjiang Province is the highest, followed by those from E'tuoke, Chifeng, and Hangjin Banner in Inner Mongolia.

Although the licorice has been successfully cultivated in many areas of China, how to increase GA contents of licorice is still an unsolved problem. Most of researches focused on successfully cultivating licorice, and less attention was paid to the suitable areas for licorice growth. As a result most of the cultivated licorice has poor quality. By our study, it can be concluded that the selection of cultivated areas is an essential task for improving the quality of licorice. The production base of licorice should be established in the original growing areas.

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